(12) INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(19) World Intellectual Property Organization
International Bureau

(43) International Publication Date
19 June 2003 (19.06.2003)

PCT

(10) International Publication Number
WO 03/050129 A1

(51) International Patent Classification[7]: C07F 9/6561, A61K 31/675

(21) International Application Number: PCT/US02/33641

(22) International Filing Date:
15 November 2002 (15.11.2002)

(25) Filing Language: English

(26) Publication Language: English

(30) Priority Data:

60/338,242	7 December 2001 (07.12.2001)	US
60/372,013	11 April 2002 (11.04.2002)	US
60/381,123	16 May 2002 (16.05.2002)	US

(71) Applicant *(for all designated States except US)*: **ELI LILLY AND COMPANY** [US/US]; Lilly Corporate Center, Indianapolis, IN 46285 (US).

(72) Inventor; and

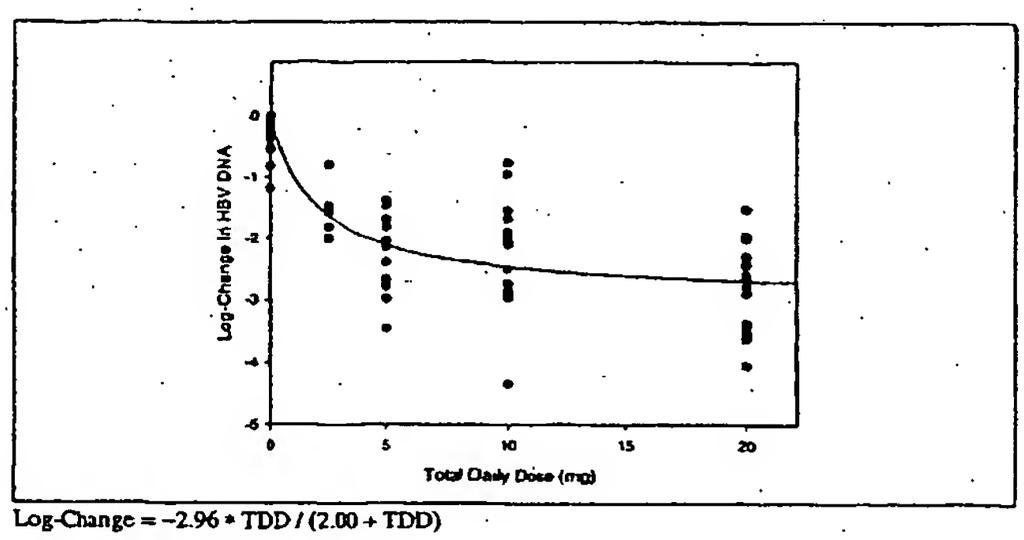
(75) Inventor/Applicant *(for US only)*: **WISE, Stephen, Douglas** [GB/SG]; Lilly-NUS Centre for Clinical Pharmacology, L6, Clinical Research Ctr (MD11), National University of Singapore, 10 Medical Drive, 117597 Singapore (SG).

(74) Agents: **COHEN, Charles, E.**, et al.; Eli Lilly And Company, P. O. Box 6288, Indianapolis, IN 46206-6288 (US).

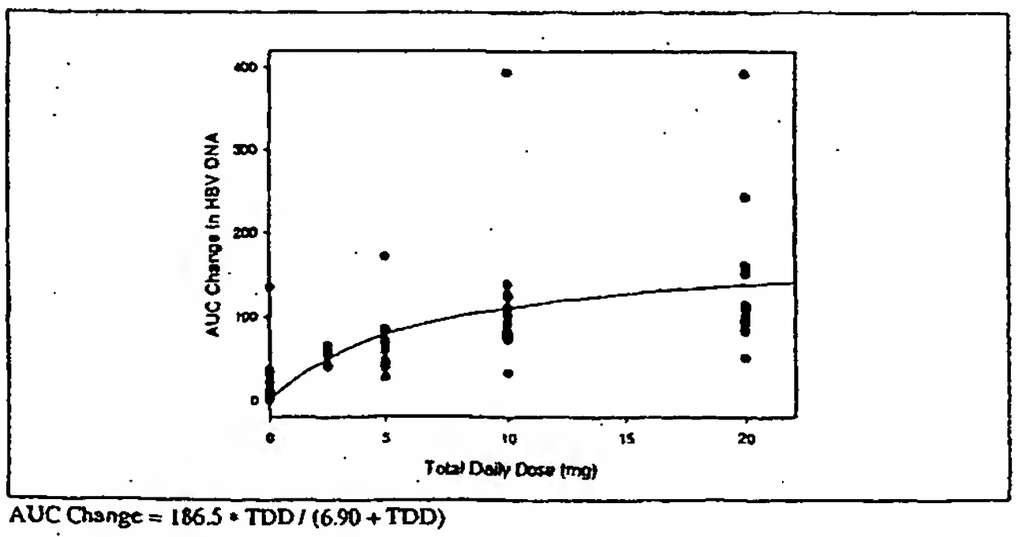
(81) Designated States *(national)*: AE, AG, AL, AM, AT (utility model), AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ (utility model), CZ, DE (utility model), DE, DK (utility model), DK, DM, DZ, EC, EE (utility model), EE, ES, FI (utility model), FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SC, SD, SE, SG, SI, SK (utility model), SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW.

[Continued on next page]

(54) Title: USE OF PHOSPHONATE NUCLEOTIDE ANALOGUE FOR TREATING HEPATITIS B VIRUS INFECTIONS



Log-Change = −2.96 * TDD / (2.00 + TDD)



AUC Change = 186.5 * TDD / (6.90 + TDD)

(57) Abstract: Provided are pharmaceutically acceptable compositions comprising 2-amino-9-[2-[bis(2,2,2-trifluoroethoxy)phosphonyl-methoxy]ethyl]-6-(4-methoxyphenylthio) purine (LY582563) for oral administration to treat hepatitis B virus infections in infected patients. The compositions are in unit dosage form including, per unit dosage, about 2.5 to about 20 mg of the purine. The compositions are adapted for oral administration, preferably in the form of a tablet or capsule, and can be administered in single or multiple doses, provided that the total daily dose of the purine is in the range of from about 2.5 mg to about 20 mg per patient per day. These compositions are particularly advantageous for lowering the plasma HBV DNA levels, or ameliorating symptoms, conditions, or disorders caused by hepatitis B virus, of human patients with chronic HBV infection. X-15484PCT331

WO 03/050129 A1

(84) **Designated States** *(regional)*: ARIPO patent (GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, SK, TR), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).

Published:
— *with international search report*

For two-letter codes and other abbreviations; refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.

USE OF PHOSPHONATE NUCLEOTIDE ANALOGUE FOR TREATING HEPATITIS B VIRUS INFECTIONS

This application claims the benefit of priority of United States Provisional Patent Applications Serial Nos. 60/338,242, filed December 7, 2001; 60/372,013, filed April 11, 2002; and 60/381,123, filed May 16, 2002, the contents of each of which are herein incorporated by reference in their entirety.

BACKGROUND OF THE INVENTION

Field of the Invention

The present invention relates to novel doses and dosing regimens for the administration of 2-amino-9-[2-[bis(2,2,2-trifluoroethoxy)phosphonylmethoxy]ethyl]-6-(4-methoxyphenylthio) purine (LY582563) to treat hepatitis B virus (HBV) infections in human patients. The invention is particularly advantageous for lowering the plasma HBV DNA levels of patients infected with HBV, and ameliorating symptoms, conditions, or disorders associated with HBV infections in humans.

Description of Related Art

Hepatitis B is a potentially fatal liver disease caused by infection with hepatitis B virus, a partially double-stranded DNA virus of the Hepadnaviridae family. It is estimated that some 350 million individuals in the world are chronically infected with HBV, and about 1 million individuals die annually as a direct result of HBV-induced cirrhosis or liver cancer.

HBV is a hepatotropic virus that replicates in the liver, resulting in acute and chronic liver disease including liver fibrosis, cirrhosis, inflammatory liver disease, and hepatic cancer that can lead to death in some patients (W.K. Joklik, *Virology*, Third Edition, Appleton & Lange, Norwalk, Connecticut, 1988). The virus is a blood-borne pathogen, which is transmitted by exposure to infectious body fluids in a fashion similar to human immunodeficiency virus (HIV). However, HBV is much more infectious than HIV. Although effective vaccines are available, they have no therapeutic value for those already infected with the virus. Estimates are that 15 to 20% of individuals who acquire the chronic infection develop cirrhosis or another chronic disability from HBV infection. For patients with compensated cirrhosis, the

survival is 84% at five years and 68% at ten years. In carriers with decompensated cirrhosis, five year survival is only 14% (Lok et al. (2001) *Hepatology* 34:1225-1241).

Alpha-interferon therapy has been available for the treatment of chronic hepatitis B for some time. However, it is effective in fewer than 40% of patients, and has dose-limiting side effects such as flu-like symptoms, weight loss, depression, and cytopenias. Lamivudine and adefovir are new compounds currently in development for use in HBV treatment. Unfortunately, the emergence of lamivudine-resistant viruses developing in up to 70% of patients after four years has been observed. It is therefore necessary and of high priority to find improved and effective anti-HBV anti-hepatitis therapies (Locarnini et. al. (1996) *Antiviral Chemistry & Chemotherapy* 7(2): 53-64).

LY582563 (2-amino-9-[2-[bis(2,2,2-trifluoroethoxy)phosphonyl-methoxy]ethyl]-6-(4-methoxyphenylthio) purine) is a recently discovered phosphonate nucleotide analog (see U. S. Patent No. 5,840,716) currently under development as an anti-HBV agent. LY582563 is a derivative of 9-[2-(phosphonyl-methoxy)ethyl]adenine (PMEA; adefovir) with improved oral absorption and antiviral activity. *In vitro*, LY582563 has demonstrated greater activity against HBV than lamivudine or PMEA. Although some of the beneficial effects of LY582563 in the treatment of HBV infections are known, optimization of treatment with this agent would be of significant therapeutic value.

SUMMARY OF THE INVENTION

Accordingly, the present invention is directed to novel pharmaceutical compositions comprising LY582563, as well as novel methods of treating HBV infections in humans, including optimal dosing regimens for the administration of LY582563 to treat HBV infected patients.

Thus, in a first aspect, the present invention provides a pharmaceutical composition for oral administration in dosage unit form, comprising:

about 2.5 mg to about 20 mg of 2-amino-9-[2-[bis(2,2,2-trifluoroethoxy)phosphonylmethoxy]ethyl]-6-(4-methoxyphenylthio) purine per dosage unit, and

a pharmaceutically acceptable carrier, diluent, or excipient.

The composition can be in the form of a tablet or capsule containing the purine in an amount of about 2.5 mg, about 5 mg, about 10 mg, about 15 mg, or about 20 mg per dosage unit, or in an amount in the range between about 5 mg and about 10 mg, or between about 7.5 mg and about 10 mg, per dosage unit. In each case, the upper limit of these ranges may be extended to about 12 or about 12.5 mg per dosage unit.

In another aspect, the present invention provides the use of 2-amino-9-[2-[bis(2,2,2-trifluoroethoxy)phosphonylmethoxy]ethyl]-6-(4-methoxyphenylthio) purine for the preparation of a medicament for treating a human patient suffering from a hepatitis B virus infection, wherein:

the medicament is formulated for oral administration, and

the medicament is in dosage unit form and comprises, per dosage unit, about 2.5 mg to about 20 mg of this purine.

The medicament can be in the form of a tablet or capsule containing the purine in an amount of about 2.5 mg, about 5 mg, about 10 mg, about 15 mg, or about 20 mg per dosage unit, or in an amount in the range between about 5 mg and about 10 mg, or between about 7.5 mg and about 10 mg, per dosage unit. In each case, the upper limit of these ranges may be extended to about 12 or about 12.5 mg per dosage unit.

In another aspect, the present invention provides a method of treating a human patient suffering from a hepatitis B virus infection, comprising administering to said patient a total amount of 2-amino-9-[2-[bis(2,2,2-trifluoroethoxy)phosphonyl-methoxy]ethyl]-6-(4-methoxyphenylthio) purine in the range of from about 2.5 mg to about 20 mg of the purine per day.

In this method, the purine can be administered to the patient for a period of time sufficient to lower the plasma level of HBV DNA of the patient compared to the plasma level of HBV DNA of the patient prior to administering the purine. Preferably, the plasma level of HBV DNA of said patient is lowered to at least about 10^4 copies/mL compared to the plasma level of HBV DNA of the patient prior to administering the purine. Alternatively, the purine can be administered to the patient for a period of time sufficient to treat or ameliorate a symptom, condition, or disorder caused by a hepatitis B virus in the patient, for example liver fibrosis, cirrhosis, inflammatory liver disease, or hepatic cancer.

In any of the foregoing methods, the purine can be administered to the patient in the form of a pharmaceutically acceptable oral composition, which can be a tablet or capsule. Furthermore, the period of time over which the purine is administered can

be several days, several weeks, several months, or several years, and the purine can be administered to the patient in a total amount of about 2.5 mg, about 5 mg, about 10 mg, about 15 mg, or about 20 mg per day, or in an amount in the range between about 5 mg and about 10 mg per day, or between about 7.5 mg and about 10 mg per day. In each case, the upper limit of these ranges may be extended to about 12 or about 12.5 mg per day. These amounts of purine can be administered in a single dose, or in divided subdoses totaling the total amount per day.

Further scope of the applicability of the present invention will become apparent from the detailed description provided below. However, it should be understood that the detailed description and specific examples, while indicating preferred embodiments of the present invention, are given by way of illustration only since various changes and modifications within the spirit and scope of the invention will become apparent to those skilled in the art from this detailed description.

BRIEF DESCRIPTION OF THE DRAWINGS

The above and other aspects, features, and advantages of the present invention will be better understood from the following detailed description taken in conjunction with the accompanying drawings, all of which are given by way of illustration only, and are not limitative of the present invention, in which:

Figure 1 shows the Profile of Mean Log-change in HBV DNA with standard deviation bars: (BID groups).

Figure 2 shows the Profile of Mean Log-change in HBV DNA with standard deviation bars: (QD groups).

Figure 3 is a scatter plot showing the Log-Change in HBV DNA on Day 29 with mean and standard deviation bars. The individual open symbols, i.e., squares, triangles, etc., represent individual patient data points. These differ to show some differences between the columns of data at different daily doses, although this is not really necessary as the columns are clearly differentiated. The solid symbol in each column represents the mean. The outer limits of the bars represent the standard deviation from that mean.

Figure 4 shows the Log-Change in HBV DNA on Day 29 with Fitted E_{max} model based on Total Daily Dose (TDD).

Figure 5 shows the Maximum Log-Change in HBV DNA during treatment period with fitted E_{max} model based on Total Daily Dose (TDD).

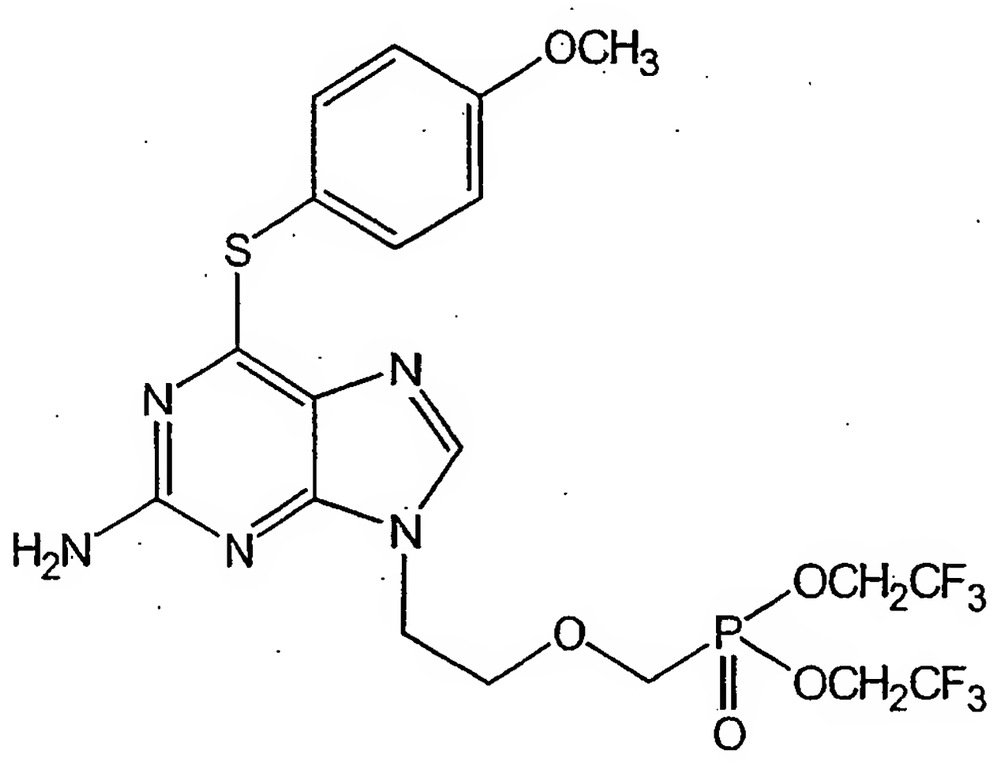
Figure 6 shows the area under the curve (AUC) of Log-Change in HBV DNA after first dose with fitted E_{max} model based on Total Daily Dose (TDD).

DETAILED DESCRIPTION OF THE INVENTION

The following detailed description of the invention is provided to aid those skilled in the in practicing the present invention. Even so, the following detailed description should not be construed to unduly limit the present invention as modifications and variations in the embodiments discussed herein can be made by those of ordinary skill in the art without departing from the spirit or scope of the present inventive discovery.

The contents of each of the references cited herein are herein incorporated by reference in their entirety.

Current treatment choices for chronic HBV infection are limited in number and clinical utility due to issues of tolerability and the emergence of resistant viral variants. LY582563, the structure of which is shown below, is a new purine nucleotide analogue prodrug that has shown potent activity against HBV in preclinical studies (Ono-Nita et. al. (2002) *Antimicrob. Agents Chemother.* 46(8): 2602-5; Wise et. al.(2002) *J. Gastroenterol. Hepatol.* 17(suppl): A46).



LY582563

For oral administration of LY582563, U.S. Patent 5,840,716 teaches that the clinical dose may generally be 0.1 mg to 500 mg/kg per day, preferably 1 mg to 50 mg/kg per day, of the compound for an adult. For an adult weighing 68 kg (approximately 150 lbs), these ranges are approximately 6.8 mg to approximately 34 g per day, preferably approximately 68 mg to approximately 3.4 g per day. The '716 patent notes that these doses may be appropriately increased or decreased depending on age, conditions or symptoms, or the presence or absence of a co-administered drug. In addition, U.S. Patent 5,840,716 teaches that the above daily doses may be administered once a day, or dividedly administered twice or several times a day with appropriate intervals. Finally, continual administration may be carried out at intervals of several days.

Determination of a safe and effective dose of any new pharmaceutical agent, and an appropriate treatment regimen employing the drug, is an empirical process. This process generally requires treating afflicted patients with varying doses of the agent, one or more times daily, over varying time periods, and monitoring effectiveness by accepted diagnostic methods. In the case of HBV, such methods can include measurement of plasma or tissue levels of HBV DNA or various antigens that comprise part of the virus, or patient response such as antibody formation (for example hepatitis B e antibody) or biochemical improvement such as decreased alanine aminotransferase (ALT) levels. In addition, one can also measure the effectiveness of treatment by monitoring the amelioration of symptoms, conditions, or disorders caused by the virus. In the case of HBV, these include fatigue, anorexia, jaundice, liver fibrosis and inflammation, cirrhosis, and hepatic cancer (hepatocellular carcinoma). In addition to demonstrating efficacy, however, one must also demonstrate acceptable safety, which includes acceptable toxicity. Toxicities that have been observed with other HBV antivirals include mitochondrial toxicity, renal toxicity, and bone marrow toxicity. The relationship between dose and treatment regimen for a new drug entity, and effectiveness and safety, can only be determined by clinical testing in humans. Therefore, determination of dose and treatment regimen parameters cannot be predicted or deduced, and the results are necessarily novel and unobvious. Particular attention must be paid to selecting treatment parameters that, while being effective to treat a disease, do not cause other harm to the patient. Given these considerations, the findings disclosed herein relating to the use of LY582563 in treating HBV infections in human patients are surprising, and could

not have been predicted before the present studies were conducted. These results provide a rational basis for safe and effective dosing and treatment regimens employing LY582563 in the treatment of HBV.

The studies disclosed herein were undertaken to determine optimal doses and dosing regimens for treating HBV-infected patients with LY582563, thereby maximizing the safety and therapeutic benefits obtainable with this antiviral pharmaceutical agent. Briefly, patients with chronic HBV infection were treated with LY582563 in a randomized, placebo controlled, dose escalating study over 28 days. Doses of LY582563 included 2.5 mg, 5 mg, and 10 mg QD or BID, and 20 mg QD. A placebo was also employed. HBV DNA in treated patients was measured during dosing and for up to twelve weeks of follow-up. Safety data, e.g., clinical signs (e.g., blood pressure, pulse, respiratory rate, and general physical findings), adverse events (e.g., upper abdominal pain, diarrhea, nausea, headache, fatigue, and alopecia), ECGs, hematology, biochemistry measures in blood (e.g., aspartate aminotransferase (AST), alanine aminotransferase (ALT), lactic acid, blood urea nitrogen (BUN), creatinine, and bilirubin), as well as highly sensitive urinary markers of renal tubular toxicity (lactate dehydrogenase (LDH), urinary creatinine, and urinary beta-N-acetyl glucosaminidase (beta-NAG)), were also measured. The data obtained demonstrate that LY582563 exhibits a favorable safety profile and effective HBV antiviral activity in chronically infected patients, and suggest optimal dose levels and dosing regimes for long term safe and effective use of this new HBV antiviral agent. Based on the present results, an appropriate dose of LY582563 balancing both safety and efficacy is between about 2.5 mg and about 20 mg per patient per day. These doses may have to be administered to patients over the course of several days, several weeks, or several months or years in order to ameliorate or control undesirable pathogenic consequences of HBV infection.

Dosing Regimen for the Treatment of HBV with LY582563

The oral dosing regimen for treating a patient suffering from HBV with LY582563 is generally selected in accordance with a variety of factors, including the age, weight, sex, diet, and medical condition of the patient, genotype of the infecting virus, the severity of the infection, and pharmacological considerations such as the activity, efficacy, pharmacokinetic, and toxicology profiles of LY582563. Administration of LY582563 should generally be continued over a period of several

hours, days, or weeks to several months or years until virus titers reach acceptable levels, or until one or more indicia, symptoms, conditions, or disorders present in a patient due to hepatitis B virus infection has been ameliorated or completely eliminated, indicating that infection has been controlled or eradicated.

With respect to reduction in plasma HBV DNA levels, the long term goal is currently a matter of debate, with some workers in the field proposing that a desirable level is below 10^4 copies/mL. The duration for this level of HBV DNA has yet to be defined.

Representative indicia of HBV infection other than plasma HBV DNA levels include, but are not limited to, liver fibrosis, cirrhosis, inflammatory liver disease, and hepatic cancer. See Hollinger et al. (2001) in *Fields Virology*, Fourth Ed., Vol. 2, David M. Knipe et al., Eds., "Hepatitis B Virus," Chapter 87, pp. 2971-3036, Lippincott, Williams, & Wilkins, Philadelphia, PA for a review. In most parts of the world, liver fibrosis, etc., is diagnosed via liver biopsy; in most Asian countries, liver biopsies are not routinely performed, and medical practitioners usually rely on clinical measures to make presumptive diagnoses.

Patients undergoing treatment with LY582563 can be routinely monitored by measuring HBV DNA levels in their serum by, for example, slot-blot, dot-blot, or PCR techniques, or by measurement of HBV antigens, such as HBV surface antigen (HBsAg) and HBV e antigen (HBeAg), in serum and tissue to determine the effectiveness of therapy. Methods therefor are discussed in Hoofnagle et al. (1997) *New Engl. Jour. Med.* 336(5):347-356; F.D. Hollinger (1996) in *Fields Virology*, Third Ed., Vol. 2, Bernard N. Fields et al., Eds., "Hepatitis B Virus," Chapter 86, pp. 2738-2807, Lippincott-Raven, Philadelphia, PA; and Hollinger et al. (2001) in *Fields Virology*, Fourth Ed., Vol. 2, David M. Knipe et al., Eds., "Hepatitis BVirus," Chapter 87, pp. 2971-3036, especially pages 2989-2991, Lippincott, Williams, & Wilkins, Philadelphia, PA, and the references cited therein. In chronic hepatitis B, remissions are characterized by the disappearance of HBV viral DNA, i.e., reduction to undetectable levels as measured by, for example, hybridization tests capable of detecting levels $\geq 10^5$ genomes per ml of serum, or HBeAg from serum despite the continued presence of HBsAg. These serologic events are followed by improvement in the biochemical and histologic features of the disease. The end point of successful treatment in most trials of antiviral therapy is the disappearance of HBeAg and viral DNA from serum. In patients in whom the e antigen disappears, remission is usually

sustained, and results in an inactive HBsAg carrier state. Many caucasian patients, most of whom acquire the disease in adulthood and receive interferon therapy, eventually become HBsAg-negative (see Hoofnagle et al. (1997) *New Engl. Jour. Med.* 336 (5) :347-356 for a review). However, this is not representative of about 90% of the world's HBV population. Continuous analysis of the data obtained by such methods permits modification of the treatment regimen during therapy so that an optimal amount of LY582563 is administered, and so that the duration of treatment can be determined as well. Thus, the treatment regimen/dosing schedule can be rationally modified over the course of therapy so that the lowest amount of LY582563 which exhibits satisfactory antiviral effectiveness is administered, and so that administration is continued only so long as is necessary to successfully treat the infection.

In the context of the present invention, the terms "ameliorate," "treat," "treatment," "therapeutic use," or "treatment regimen" as used herein are meant to encompass prophylactic, palliative, and therapeutic modalities of administration of LY582563, and include any and all uses of this purine that remedy an indicium, a disease state, a condition, a symptom, or a disorder caused by a hepatitis B virus, or which prevent, hinder, retard, or reverse the progression of indicia, symptoms, conditions, or disorders associated with HBV infection. Thus, any prevention, improvement, alleviation, reversal, or complete elimination of an undesirable indicium, disease state, symptom, condition, or disorder associated with HBV infection is encompassed by the present invention. The term "dosage unit" as used herein refers to an individual delivery vehicle, for example, but not limited to, a tablet or capsule, for administration of a dose of the active LY582563 purine.

The following example is provided to illustrate various aspects of the present invention, and should not be construed to be limiting thereof in any way.

Example 1

Multiple Dose-Escalation Study

in Healthy Subjects and Patients with Chronic Hepatitis B Infection

A randomized, single period, multiple dose-escalation, single-blind study of LY582563 was carried out in 2 parts. Part 1 was conducted in healthy subjects, and Part 2 was conducted in patients with compensated chronic HBV infection (hereafter

"patients"). The study was designed to evaluate the safety and pharmacodynamics of multiple doses of LY582563. This was the first multiple dose study in humans.

Dose escalation was carried out in conservative increments in healthy subjects who were given BID regimen for a 2-week period. Safety data were reviewed prior to each dose escalation.

Part 1 (Healthy Subjects)

In part 1, there were 4 groups with 8 subjects in each group (6 subjects on study drug and 2 on placebo). Table 1 shows the characteristics of the subjects in this study. The LY582563 doses were: 2.5 mg BID, 5 mg BID, 10 mg BID and 15 mg BID. Approximately 24 hours after a single dose of LY582563, multiple BID dosing for approximately 14 days of LY582563 was administered, except for the last day of dosing which was administered QD. There was a dose escalation for each consecutive group of subjects. Safety and other data, as appropriate, were reviewed prior to each dose escalation step. Plasma concentrations of LY582563 and its metabolites (602074, 602075 and 602076) were measured up to 24 hours after a single dose of LY582563 and after the last dose by a validated LC/MS/MS method (Advion BioSciences, Inc., Ithaca, NY). Subjects were followed up at regular intervals for approximately 4 weeks after the last dose of LY582563.

Table 1. Characteristics of Healthy Subjects

	LY582563 Dose				
	2.5 mg BID	5.0 mg BID	10 mg BID	15 mg BID	Placebo
No. of Patients, *n*	6	6	6	6	8
Age (years)	23.5±1.8	23.5±2.6	23.0±1.1	22.8±1.0	23.4±2.3
Male:Female	6:0	6:0	6:0	6:0	8:0
Ethnicity, n (% Chinese)	4 (67%)	4 (67%)	6 (100%)	5 (83%)	7 (87%)
BMI, kg/m^2	21.7±1.0	20.9±1.6	25.8±1.1	23.7±2.5	24.6±2.8

Part 2 (Patients)

In part 2, there were 7 groups with 8-12 patients in each group. Table 2 shows the characteristics of the subjects in this study. The LY582563 doses were: 2.5 mg BID (n=8), 5 mg BID (n = 8), 10 mg BID (n = 8), 2.5 mg QD (n = 8), 5 mg QD (n = 11), 10 mg QD (n = 12) and 20 mg QD (n = 11). Approximately 24 hours after a

single dose of LY582563 (day 1), multiple BID dosing for approximately 28 days (i.e., day 2 to day 29) of LY582563 was administered, except for the last day (day 29) of dosing, which was QD. Patients were only studied following review of safety data from healthy subjects at the same dose level in Part 1. Patients were followed up at regular intervals for approximately 12 weeks after the last dose of LY582563. Plasma concentrations of LY582563 and its metabolites (602074, 602075 and 602076) were sampled up to 24 hours after a single dose of LY582563 and after the last dose. Subjects were followed up at regular intervals for approximately 12 weeks after the last dose of LY582563.

Table 2. Patient Characteristics

	LY582563 Dose							Placebo
	2.5 mg BID	5.0 mg BID	10 mg BID	2.5 mg QD	5.0 mg QD	10 mg QD	20 mg QD	
No. of Patients, n	6	6	6	6	7	9	9	16
Age (years)	32.5±7.7	35.7±6.0	38.8±9.0	28.0±4.6	28.4±7.6	34.9±11.5	38.4±10.2	34.2±8.8
Male:Female	6:0	6:0	5:1	6:0	7:0	9:0	9:0	14:2
Ethnicity, n (% Chinese)	5 (83%)	5 (83%)	6 (100%)	6 (100%)	6 (86%)	9 (100%)	8 (89%)	16 (100%)
BMI, kg/m^2	23.4±2.3	25.4±3.2	25.1±3.1	23.8±2.4	24.6±3.1	24.0±3.3	23.8±3.4	22.7±3.0
HBe Ag positive, n (%)	6 (100%)	6 (100%)	3 (50%)	5 (83%)	6 (86%)	5 (56%)	6 (67%)	12 (75%)
HBVDNA*(pg /mL)								
- Geometric Mean	$1.19x10^9$	$2.71x10^9$	$5.03x10^8$	$2.92x10^8$	$6.24x10^8$	$3.73x10^8$	$7.39x10^8$	$6.78x10^8$
- Range	$3.17x10^8$ - $3.69x10^9$	$7.99x10^7$ – $5.69x10^9$	$4.64x10^5$ – $2.20x10^9$	$8.02x10^6$ – $7.50x10^8$	$2.85x10^5$ – $1.85x10^9$	$2.46x10^6$ – $8.22x10^8$	$1.58x10^5$ – $2.80x10^9$	$2.95x10^5$ – $3.29x10^9$

*: HBV DNA Baseline = Geometric mean of HBV DNA at screening, -28 day, -14 day and predose.

Treatments

LY582563 and matching placebos were supplied as either 2.5 mg, 10 mg, or 20 mg tablets. LY582563 and matching placebo tablets were supplied by Mitsubishi Pharma Corporation, Tokyo, Japan.

LY582563 and placebo formulations for oral administration were as follows:

Table 3. 2.5 mg LY582563 Tablet

Ingredient	Quantity (mg/tablet)	Function
Active Ingredient		
LY582563	2.5	Active ingredient
Other Ingredients		
D-mannitol	92.5	Diluent
Corn starch	23.1	Diluent
Hydroxypropylcellulose	3.1	Binder
Low substituted hydroxypropylcellulose	1.3	Disintegrant
Magnesium stearate	2.5	Lubricant
Hydroxypropylmethyl-cellulose 2910	3.35	Coating agent
Propylene Glycol	0.77	Coating agent
Titanium Dioxide	0.33	Coating agent
Talc	0.55	Coating agent
Hydrogenated Oil	0.0625	Coating agent

5

Table 4. 10 mg LY582563 Tablet

Ingredient	Quantity (mg/tablet)	Function
Active Ingredient		
LY582563	10	Active ingredient
Other Ingredients		
D-mannitol	84.5	Diluent
Corn starch	21.1	Diluent
Hydroxypropylcellulose	3.1	Binder
Low substituted hydroxypropylcellulose	3.8	Disintegrant
Magnesium stearate	2.5	Lubricant
Hydroxypropylmethyl-cellulose 2910	3.35	Coating agent
Propylene Glycol	0:77	Coating agent
Titanium Dioxide	0.33	Coating agent
Talc	0.55	Coating agent
Hydrogenated Oil	0.0625	Coating agent

5

Table 5. 20 mg LY582563 Tablet

Ingredient	Quantity (mg/tablet)	Function
Active Ingredient[1]		
LY582563	20	Active ingredient
Other Ingredients		
D-mannitol	74.5	Diluent
Corn starch	18.6	Diluent
Hydroxypropylcellulose	3.1	Binder
Low substituted hydroxypropylcellulose	6.3	Disintegrant
Magnesium stearate	2.5	Lubricant
Hydroxypropylmethyl-cellulose 2910	3.35	Coating agent
Propylene Glycol	0.77	Coating agent
Titanium Dioxide	0.33	Coating agent
Talc	0.55	Coating agent
Hydrogenated Oil	0.0625	Coating agent

10

15

20

5

Table 6. Placebo Tablet

Ingredient	Quantity (mg/tablet)	Function
Active Ingredient[1]		
LY582563	0	Active ingredient
Other Ingredients		
D-mannitol	90.5	Diluent
Corn starch	22.6	Diluent
Hydroxypropylcellulose	3.1	Binder
Low substituted hydroxypropylcellulose	6.3	Disintegrant
Magnesium stearate	2.5	Lubricant
Hydroxypropylmethyl-cellulose 2910	3.35	Coating agent
Propylene Glycol	0.77	Coating agent
Titanium Dioxide	0.33	Coating agent
Talc	0.55	Coating agent
Hydrogenated Oil	0.0625	Coating agent

10

Method of Manufacture

LY582563, D-mannitol, and corn starch are mixed. Hydroxypropylcellulose dissolved in purified water is added for granulation by fluid-bed granulator. After drying, granules are screened by tornado mill. Screened granules are mixed with magnesium stearate and low substituted hydroxypropylcellulose by V-shaped mixer. Mixed granules are compressed into tablets. Tablets are filmed coated with hydroxypropylmethyl-cellulose 2910, propylene glycol, titanium dioxide, and talc by conventional methods. Finally, tablets are coated with a little hydrogenated oil.

Dosing

The 2.5 mg dose was administered as 1 tablet of 2.5 mg LY582563 or 1 matching placebo. The 5 mg dose was administered as 2 tablets of 2.5 mg LY582563 or 2 matching placebos. The 10 mg dose was administered as 1 tablet of 10 mg LY582563 or 1 matching placebo. The 15 mg dose was administered as 2 tablets of 2.5 mg LY582563 and 1 tablet of 10 mg LY582563 or matching placebos corresponding to the 2.5 and 10 mg LY582563 tablets. The 20 mg dose was administered as 1 tablet of 20 mg LY582563 or 1 matching placebo.

Each participant was randomly assigned to each dose group and to receive either LY582563 or placebo. Healthy subjects in Part 1 and patients in Part 2 were allocated according to a randomization block of 4 (3 study drug to one placebo). Study drug was administered BID for healthy subjects in Part 1 (Table 7) and BID or QD for patients in Part 2 (Table 8).

TABLE 7. Dosing Groups for Part 1 (Healthy Subjects)

Dose Group*	Dose of LY582563 (mg) and Dosing Schedule	n (on study drug)	n (on placebo)
1	2.5 BID	6	2
2	5 BID	6	2
3	10 BID	6	2
4	15 BID	6	2

*There was a dose escalation for each consecutive group. Safety and other data, as appropriate, were reviewed prior to each dose escalation step.

TABLE 8. Dosing Groups for Part 2 (Patients)

Dose Group*	Dose of LY582563 (mg) & Dosing Schedule	n (on study drug)	n (on placebo)
1	2.5 BID	6	2
2	5 BID	6	2
3A	2.5 QD	3	1
	10 BID	6	2
3B	2.5 QD	3	1
	10 QD	6	2
4A	20 QD	9	2
4B	10 QD	3	1
5A	5 QD	8**	3

*The order of grouping was based on the time when the group started study drug therapy. Patients were only studied following review of safety data from healthy subjects at the same or equivalent dose level in Part 1. In this study, QD dosing for patients started with 2.5 mg (3A and 3B), followed by 10 mg (3B and 4B)) and 20 mg (4A). It was decided that QD doses higher than 20 mg would not be studied following the review of safety data in healthy subjects of 15 mg BID dose. Therefore, 5 mg QD dose was studied in 5A group.

**1subject of 5A dose group who was randomized to receive LY582563 was withdrawn from the study on day 1 prior to receiving study drug.

Pharmacodynamic Assessment

Blood samples were taken from hepatitis B infected patients for the measurement of plasma concentrations of HBV DNA levels in Part 2 of the study. The level of plasma HBV DNA was determined using a validated HBV polymerase chain reaction (PCR) assay (NGI HBV SuperQuant™ Quantitative PCR Assay; National Genetics Institute, Los Angeles, CA). All HBV DNA levels were transformed to the $\log_{10}$ scale. Baseline measurements were defined as the geometric mean of the non log-transformed data obtained prior to dosing. Within-subject maximum log-changes in HBV DNA values from baseline during the treatment period were derived by substracting the minimum HBV DNA value during treatment period from the baseline value. For two patients (2108 and 3308, both on placebo treatment) where the HBV DNA values during treatment were all greater than the baseline, the maximum log-change was assigned a value of zero.

To take into consideration the rate of decline and rate of return of HBV DNA values during the study period, the area under the curve (AUC) for log change between each HBV DNA time curve and a horizontal line passing through the baseline score were computed. The HBV DNA values were first converted to baseline score when they were higher than baseline score, and area between the curve and the

horizontal baseline was computed using the trapezoidal rule to adjust for the difference in time interval between measurements.

In the analysis, three HBV DNA values that were above the specified assay range ($5x10^9$ genome copies/mL) without any dilution assays performed were assigned a value of $5x10^9$ copies/mL, and one HBV DNA value that was below the specified assay range (100 genome copies/mL) was assigned a value of 100 copies/mL

Plasma samples for HBV DNA were quantitated using the validated NGI HBV SuperQuant™ Quantitative PCR assay with a specified assay range of 100 genome copies/mL to $5x10^9$ genome copies/mL at National Genetics Institute (Los Angeles, CA).. Blood samples of approximately 5 mL were collected at screening, at approximately 4 and 2 weeks before dosing, as well as just prior to dosing; measurements were then made during the dosing period (approximately 3 days, then 1, 2, 3, and 4 weeks after the first dosing) and the follow up period (5, 6, 8, 12 and 16 weeks after the first dosing). Depending on the level of hepatitis B viral suppression 8 weeks after dosing, subjects would have additional samples taken approximately 10 and 14 weeks after the first dosing.

On dosing days, the blood samples were taken prior to the morning dose. Sampling times were adjusted, added, or deleted based on practical considerations, clinical needs, or need for additional pharmacodynamic data.

The means and standard deviations of HBV DNA values were obtained at each scheduled time point for each dosing group.

For the log-drop on day 29, maximum log-change and AUC for log-change, a regression model was used to compare the mean parameter values of the dosing groups. The dose level nested within dosing regimen was used as fixed effect, and baseline value used as covariate. Differences in the mean values were obtained with 95% confidence intervals. An exploratory E_{max} model (J. Gabrielsson and D. Weiner (1998), *Pharmacokinetic and Pharmacodynamic Data Analysis: Concepts and Applications,* Second Edition, Swedish Pharmaceutical Press) was also applied to the above parameters to obtained a fitted regression curve with total daily dose as explanatory variable.

Pharmacodynamic Evaluation

The profiles of mean log-change in HBV DNA are shown in Figures 1 and 2. The placebo group exhibited a consistent mean value of zero (i.e., not significantly different from baseline) at each measurement day. For BID dosing, the 2.5, 5, and 10 mg doses of LY582563 resulted in a linear decline in log-scale of HBV DNA during the treatment period (Figure 1). The same was also observed for QD dosing groups, although the 2.5 mg dose group appeared to have a slower rate of decline, and the 5 mg group exhibited a slower rate of decline after Day 7 (Figure 2). Generally, the HBV DNA values gradually returned to baseline after the last dose of LY582563, and the time of return to the baseline or 1/2 log change of baseline appeared to increase with higher doses. The mean HBV DNA for 10 mg BID, 10 mg QD, and 20 mg QD groups was lower compared to the baseline at day 112 (Figures 1 and 2), indicating that the viral load did not return to baseline after a 3-month period.

A scatter plot of the log-change from baseline of HBV DNA is presented in Figure 3. The pair-wise comparisons of mean log-change on day 29 within each dosing regimen are listed in Table 9. Pair-wise differences were tested at 5% level of significance without any multiplicity adjustments.

Table 9. Mean Log-Change from Baseline of HBV DNA Value with 95% Confidence Interval on Day 29

Dose (mg)	n	Mean	Difference vs. Placebo	Difference vs. 2.5mg	Difference vs. 5mg	Difference vs. 10mg
Placebo	15	0.0639				
2.5mg BID	6	2.02	1.95 (1.35, 2.56)			
5.0mg BID	5	1.98*	1.92 (1.11, 2.72)	-0.04 (-1.01, 0.94)		
10mg BID	6	2.53	2.47 (2.02, 2.92)	0.51 (-0.22, 1.25)	0.55 (-0.36, 1.46)	
2.5mg QD	6	1.52	1.46 (1.09, 1.83)			
5.0mg QD	7	1.96	1.89 (1.13, 2.66)	0.44 (-0.39, 1.26)		
10mg QD	9	2.49	2.43 (1.83, 3.03)	0.97 (0.30, 1.64)	0.54 (-0.42, 1.49)	
20mg QD	9	2.50	2.44 (1.88, 3.00)	0.98 (0.34, 1.62)	0.54 (-0.39, 1.48)	0.01 (-0.79, 0.80)

Note: Confidence interval (shown in parenthesis) that does not contain zero indicates a statistically significant difference between two treatments at the 5% level.
*One subject did not have HBV DNA data on day 29.

For BID dosing, the mean log-change on day 29 was observed to be 2.02, 1.98, and 2.53 for the 2.5, 5, and 10 mg dose groups, respectively. All mean log-changes were statistically significant when compared to those of the placebo group. No statistically significant difference was detected between the three BID dose levels. For QD dosing, the observed mean log-changes on day 29 were 1.52, 1.96, 2.49, and 2.50 for the 2.5, 5, 10, and 20 mg dose groups, respectively. All mean log-changes were statistically significant when compared to those of the placebo group. The mean log-changes for the 10 and 20 mg dose groups were similar, and were significantly different from that of 2.5 mg group.

An E_{max} model was fitted to the log-change with total daily dose as the explanatory variable (Figure 4), facilitating understanding the relationship between pharmacodynamic effects and total daily dose. The fitted regression curve suggests that a plateau was achieved for HBV DNA drop at approximately 10 mg total daily dose.

The pair-wise comparisons of mean maximum log-change in HBV DNA values during the treatment period within each dosing regimen are listed in Table 10. Pair-wise differences were tested at 5% level of significance without any multiplicity adjustments.

Table 10. Mean Maximum Log-Change from Baseline of HBV DNA Value with 95% Confidence Interval During Treatment Period (Day 1 to Day 29)

Dose (mg)	n	Mean	Difference vs. Placebo	Difference vs. 2.5mg	Difference vs. 5mg	Difference vs. 10mg
Placebo	16	0.34				
2.5mg BID	6	2.39	2.05 (1.50, 2.60)			
5.0mg BID	6	1.82	1.48 (0.75, 2.21)	-0.57 (-1.45, 0.30)		
10mg BID	6	2.93	2.59 (2.03, 3.14)	0.54 (-0.21, 1.29)	1.11 (0.22, 2.00)	
2.5mg QD	6	1.53	1.19 (0.82, 1.57)			
5.0mg QD	7	2.19	1.85 (1.33, 2.37)	0.66 (0.06, 1.25)		
10mg QD	9	2.58	2.24 (1.66, 2.81)	1.04 (0.39, 1.69)	0.39 (-0.35, 1.13)	
20mg QD	9	2.68	2.34 (1.79, 2.88)	1.14 (0.52, 1.77)	0.49 (-0.23, 1.21)	0.10 (-0.66, 0.86)

Note: Confidence interval (shown in parenthesis) that does not contain zero indicates a statistically significant difference between two treatments at the 5% level.

For BID dosing, the mean maximum log-changes during the treatment period were observed to be 2.39, 1.82, and 2.93 for the 2.5, 5, and 10 mg dose groups, respectively. All mean maximum log-drops were statistically significant when compared to that of the placebo group. Although there appeared to be a significant difference in mean log-change between the 10 mg and 5 mg dose groups, there was no significant difference between the 10 mg and 2.5 mg dose groups. For QD dosing, the observed mean maximum log-changes during the treatment period were 1.53, 2.19, 2.58, and 2.68 for the 2.5, 5, 10, and 20 mg dose groups, respectively. All mean log-drops were statistically significant when compared to that of the placebo group. The mean log-change for the 5, 10, and 20 mg dose groups appeared to be similar, and were significantly different from that of 2.5 mg group.

At 10 mg BID, 10 mg QD, and 20 mg QD, all patients demonstrated a maximum log change in HBV DNA greater than 1.5 during the treatment period.

An E_{max} model was fitted to the maximum log-change with total daily dose as the explanatory variable (Figure 5). The fitted regression curve suggests achievement of a plateau for maximum HBV DNA drop during treatment at approximately 10 mg total daily dose.

For further clarification, Figure 4 shows the log-change in HBV DNA on day 29 with fitted E_{max} model based on total daily dose (TDD); Figure 5 shows the maximum log-change in HBV DNA over the treatment period with fitted E_{max} model based on TDD. Both figures need to be taken into consideration as the maximum log-change for some patients occurred at times other than at day 29. Taken together, these figures permit a better understanding of the profile of pharmacodynamic effect, i.e., HBV DNA change, with dose.

The pair-wise comparisons of mean change in AUC after the first dose until the end of the study period (day 112) within each dosing regimen are listed in Table 11. Pair-wise differences were tested at the 5% level of significance without any multiplicity adjustments.

Table 11. Mean AUC of Log-Change from Baseline of HBV DNA Value with 95% Confidence Interval After First Dose (Day 1 to Last Visit)

Dose (mg)	n	Mean	Difference vs. Placebo	Difference vs. 2.5mg	Difference vs. 5mg	Difference vs. 10mg
Placebo	15	18.4				
2.5mg BID	6	76.2	57.7 (31.9, 83.6)			
5.0mg BID	5	91.6	73.1 (34.9, 111)	15.4 (-24.0, 54.7)		
10mg BID	6	135	117 (71.7, 162)	59.1 (11.2, 107)	43.7 (-12.0, 99.4)	
2.5mg QD	6	53.4	34.9 (18.0, 51.9)			
5.0mg QD	7	75.2	56.7 (26.4, 87.0)	21.8 (-5.33, 48.9)		
10mg QD	9	142	123 (58.7, 188)	88.5 (25.1, 152)	66.7 (-1.43, 135)	
20mg QD	9	125	106 (35.5, 177)	71.1 (1.84, 140)	49.4 (-24.4, 123)	-17.4 (-111, 76.0)

Note: Confidence interval (shown in parenthesis) that does not contain zero indicates a statistically significant difference between two treatments at the 5% level.

For BID dosing, the mean AUC for log-change after the first dose for the 2.5, 5, and 10 mg dose groups was observed to be statistically significant when compared to that of the placebo group. A significant difference in mean AUC for log-change between the 10 mg and 2.5 mg dose groups was also observed. For QD dosing, the observed mean AUC for log-change for the 2.5, 5, 10, and 20 mg dose groups was statistically significant compared to that of the placebo group. The mean AUC for log-change for the 10 and 20 mg dose groups appeared to be similar, and were significantly different from that of the 2.5 mg group. A trend of significant difference between the 10 mg and 5 mg doses was also observed.

An E_{max} model was fitted to the AUC for log-change with total daily dose as the explanatory variable, similar to the E_{max} model for log-change in HBV DNA (Figure 6). The fitted regression curve suggests that a 10 mg total daily dose would achieve 80% of the 20 mg total daily dose's effect on AUC log-change in HBV DNA.

Conclusions Regarding Efficacy of LY582563

As demonstrated by the data presented above, all doses of LY582563 tested in this study were associated with a reduction of plasma HBV DNA during the treatment period, and the reduction appeared to be dose-dependent. The 10 mg QD, 10 mg BID, and 20 mg QD dose groups showed the greatest viral suppression, as seen in the mean

drop of HBV DNA on day 29 (Figure 3), the mean maximum log-viral decline (Table 10), and the mean AUC change in log-viral decline after first dosing (Table 11). This level of reduction achieved with agents such as interferon alpha, lamivudine, and adefovir has been demonstrated to continue to progress to further depression of HBV DNA, with a cumulative increase in seroconversion with duration of therapy. The data suggest a plateau of effect before 10 mg total daily dose. A dose in the range between about 5 mg per day and about 10 mg per day, or between about 7.5 mg per day and about 10 mg per day, may be optimal. In each case, the upper limit of these ranges may be extended to about 12 or about 12.5 mg per day. These doses can be administered in tablet or capsule form.

The viral load gradually returned to baseline after cessation of LY582563 treatment, with the higher dose groups demonstrating a slower recovery of viral load (Figures 1 and 2). Slower return of viral replication in the higher dose groups could be due to a greater extent of viral suppression during treatment, thus leading to a smaller remnant pool of infected hepatocytes. As the viral load at day 29 was different in each dose group, with higher dose groups having a lower viral load compared to the lower dose groups that had a higher viral load (Figures 1 and 2), no analysis was performed to determine the return of viral load toward baseline after the cessation of LY582563 treatment. This study was not designed to detect the mean log-change from baseline of HBV DNA value on day 29 between dose groups.

Safety of LY582563

The most commonly reported adverse events in healthy subjects were somnolence, upper abdominal pain, and diarrhea. In patients with chronic hepatitis B infection, the most commonly reported adverse events were somnolence, headache, and intravenous cannula related events. There did not appear to be a dose relationship to adverse events, with the highest dose levels (10 mg BID, 20 mg QD) having an adverse event incidence comparable to or less than that of placebo. No subject was withdrawn from the study because of adverse events.

There appeared to be a dose relationship to ALT elevations, with higher dose groups having the highest prevalence of ALT increases from baseline, in both healthy subjects (approximately 30%) and patients with compensated chronic HBV infection (approximately 50-70%). The maximum extent of ALT elevations attributable to LY582563 was approximately three times the upper limit of normal.

Evaluations of clinical biochemical markers (serum phosphate, urea, and creatinine) as well as of clinical urinary markers (LDH/creatinine and beta-NAG/creatinine ratios) in both healthy subjects and patients did not reveal any dose-related renal tubular toxicity. Clinical laboratory results of plasma lactate and serum creatine kinase also suggested that there was no dose-related mitochondrial or musculoskeletal toxicity following repeated dosing with the doses of LY582563 examined. There were no clinically significant abnormalities of vital signs, electrocardiogram (ECG), or QT_c prolongations of greater than 30 msec in either healthy subjects or patients given LY582563.

The results presented above demonstrate that LY582563 was safe and well tolerated when administered as multiple doses of 2.5 mg to 30 mg daily for up to 14 days in healthy subjects, and as multiple doses of 2.5 mg to 20 mg daily for up to 28 days in patients with compensated chronic HBV infection. There appeared to be a dose relationship to ALT elevations, with higher dose groups having the highest prevalence of ALT increases from baseline, in both healthy subjects and patients with compensated chronic HBV infection. The maximum extent of ALT elevations attributable to LY582563 was approximately three times the upper limit of normal. No other biochemical markers exhibited any relationship to dose. There were no significant abnormalities of vital signs or ECGs (including assessment for QT_c prolongations) in healthy subjects and patients with compensated chronic HBV infection. All doses of LY582563 were associated with a reduction in viral load (plasma HBV DNA) during the treatment period that appeared to be dose-related. The greatest decline was observed in the 10 mg QD, 10 mg BID, and 20 mg QD dose groups, resulting in an approximately 2.5 mean $\log_{10}$ decline after four weeks of therapy. The viral load gradually returned to baseline after cessation of LY582563 treatment. This appeared to be delayed in the higher doses. Based on these results, an appropriate dose of LY582563 that balances safety with efficacy is between about 2.5 mg and about 20 mg per patient per day. These doses may have to be administered to patients over the course of several days, several weeks, or several months or years in order to ameliorate or control undesirable pathogenic consequences of HBV infection.

The invention being thus described, it is obvious that the same can be varied in many ways. Such variations are not to be regarded as a departure from the spirit and scope of the present invention, and all such modifications as would be obvious to one skilled in the art are intended to be included within the scope of the following claims.

WHAT IS CLAIMED IS:

1. A pharmaceutical composition for oral administration in dosage unit form, comprising:

about 2.5 mg to about 20 mg of 2-amino-9-[2-[bis(2,2,2-trifluoroethoxy)phosphonylmethoxy]ethyl]-6-(4-methoxyphenylthio) purine per dosage unit, and

a pharmaceutically acceptable carrier, diluent, or excipient.

2. The composition of claim 1, which is in the form of a tablet or capsule.

3. The composition of claim 1 or 2, wherein said 2-amino-9-[2-[bis(2,2,2-trifluoroethoxy)phosphonylmethoxy]ethyl]-6-(4-methoxyphenylthio) purine is present in an amount of about 2.5 mg, about 5 mg, about 10 mg, about 15 mg, or about 20 mg per dosage unit.

4. Use of 2-amino-9-[2-[bis(2,2,2-trifluoroethoxy)phosphonylmethoxy]ethyl]-6-(4-methoxyphenylthio) purine for the preparation of a medicament for treating a human patient suffering from a hepatitis B virus infection, wherein:

said medicament is formulated for oral administration, and

said medicament is in dosage unit form and comprises, per dosage unit, about 2.5 mg to about 20 mg of said purine.

5. The use according to claim 4, wherein said medicament is in the form of a tablet or capsule.

6. The use according to claim 4 or 5, wherein said 2-amino-9-[2-[bis(2,2,2-trifluoroethoxy)phosphonylmethoxy]ethyl]-6-(4-methoxyphenylthio) purine is present in an amount of about 2.5 mg, about 5 mg, about 10 mg, about 15 mg, or about 20 mg per dosage unit.

7. A method of treating a human patient suffering from a hepatitis B virus infection, comprising administering to said patient a total amount of 2-amino-9-[2-

[bis(2,2,2-trifluoroethoxy)phosphonylmethoxy]ethyl]-6-(4-methoxyphenylthio) purine in the range of from about 2.5 mg to about 20 mg of said purine per day.

8. The method of claim 7, wherein said 2-amino-9-[2-[bis(2,2,2-trifluoroethoxy)phosphonylmethoxy]ethyl]-6-(4-methoxyphenylthio) purine is administered to said patient for a period of time sufficient to lower the plasma level of HBV DNA of said patient compared to the plasma level of HBV DNA of said patient prior to administering said purine.

9. The method of claim 8, wherein said plasma level of HBV DNA of said patient is lowered to at least about 10^4 copies/mL compared to the plasma level of HBV DNA of said patient prior to administering said purine.

10. The method of claim 7, wherein said 2-amino-9-[2-[bis(2,2,2-trifluoroethoxy)phosphonylmethoxy]ethyl]-6-(4-methoxyphenylthio) purine is administered to said patient for a period of time sufficient to ameliorate an indicium, a symptom, a condition, or a disorder caused by said hepatitis B virus in said patient.

11. The method of claim 10, wherein said indicium, symptom, condition, or disorder is selected from the group consisting of liver fibrosis, cirrhosis, inflammatory liver disease, and hepatic cancer.

12. The method of any one of claims 7 to 11, wherein said 2-amino-9-[2-[bis(2,2,2-trifluoroethoxy)phosphonylmethoxy]ethyl]-6-(4-methoxyphenylthio) purine is administered to said patient in the form of a pharmaceutically acceptable oral composition.

13. The method of any one of claims 7 to 12, wherein said pharmaceutically acceptable oral composition is in the form of a tablet or capsule.

14. The method of any one of claims 8 to 13, wherein said period of time is several days, several weeks, several months, or several years.

15. The method of any one of claims 7 to 14, wherein said 2-amino-9-[2-[bis(2,2,2-trifluoroethoxy)phosphonylmethoxy]ethyl]-6-(4-methoxyphenylthio) purine is administered to said patient in a total amount of about 2.5 mg, about 5 mg, about 10 mg, about 15 mg, or about 20 mg per day.

16. The method of any one of claims 7 to 15, wherein said total amount of said 2-amino-9-[2-[bis(2,2,2-trifluoroethoxy)phosphonylmethoxy]ethyl]-6-(4-methoxyphenylthio) purine is administered in a single dose, or in divided subdoses totaling said total amount per day.

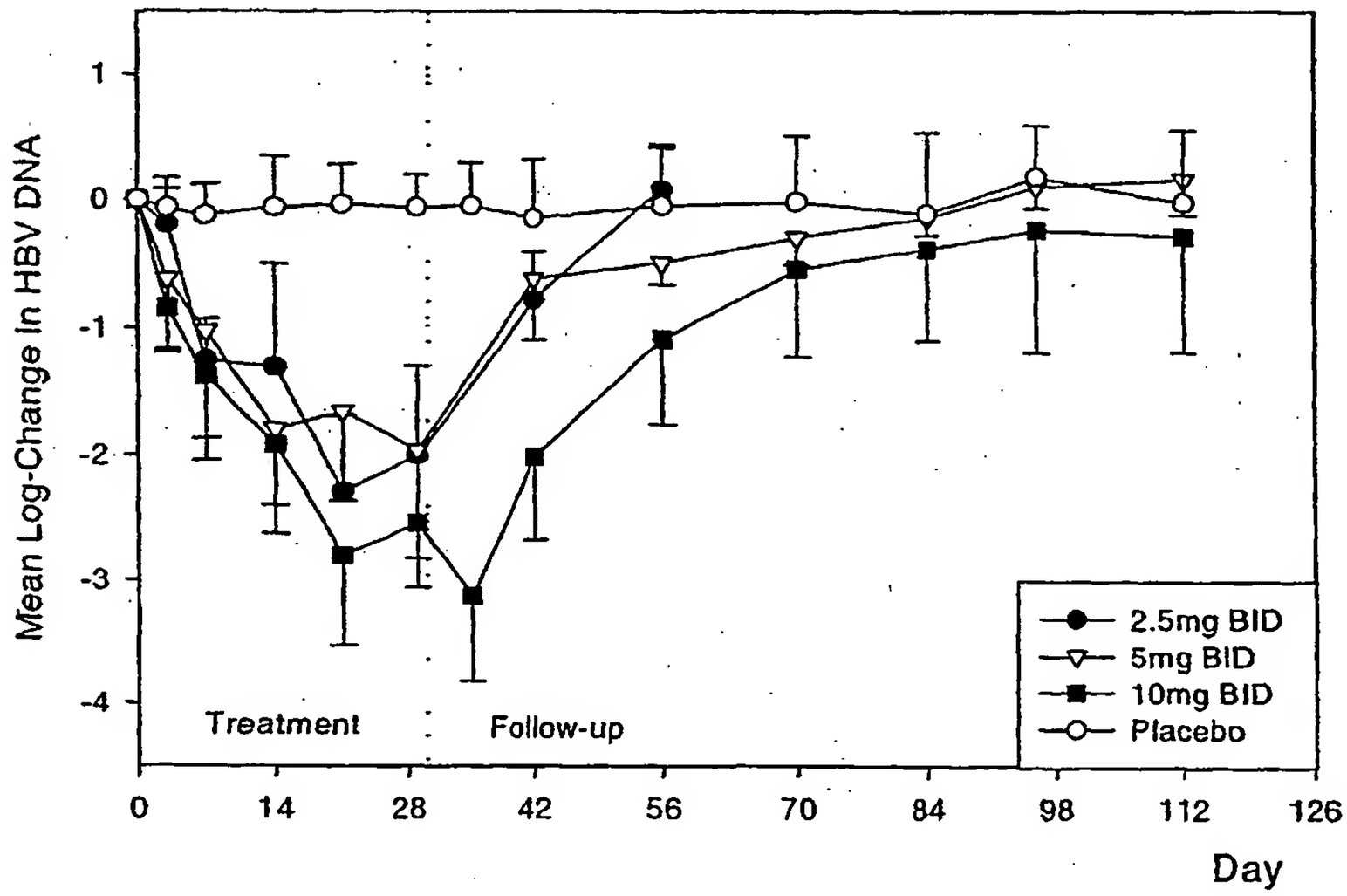

Mean Log-Change In HBV DNA
1
0
-1
-2
-3
-4
Treatment
Follow-up
2.5mg BID
5mg BID
10mg BID
Placebo
0
14
28
42
56
70
84
98
112
126
Day

FIGURE 1

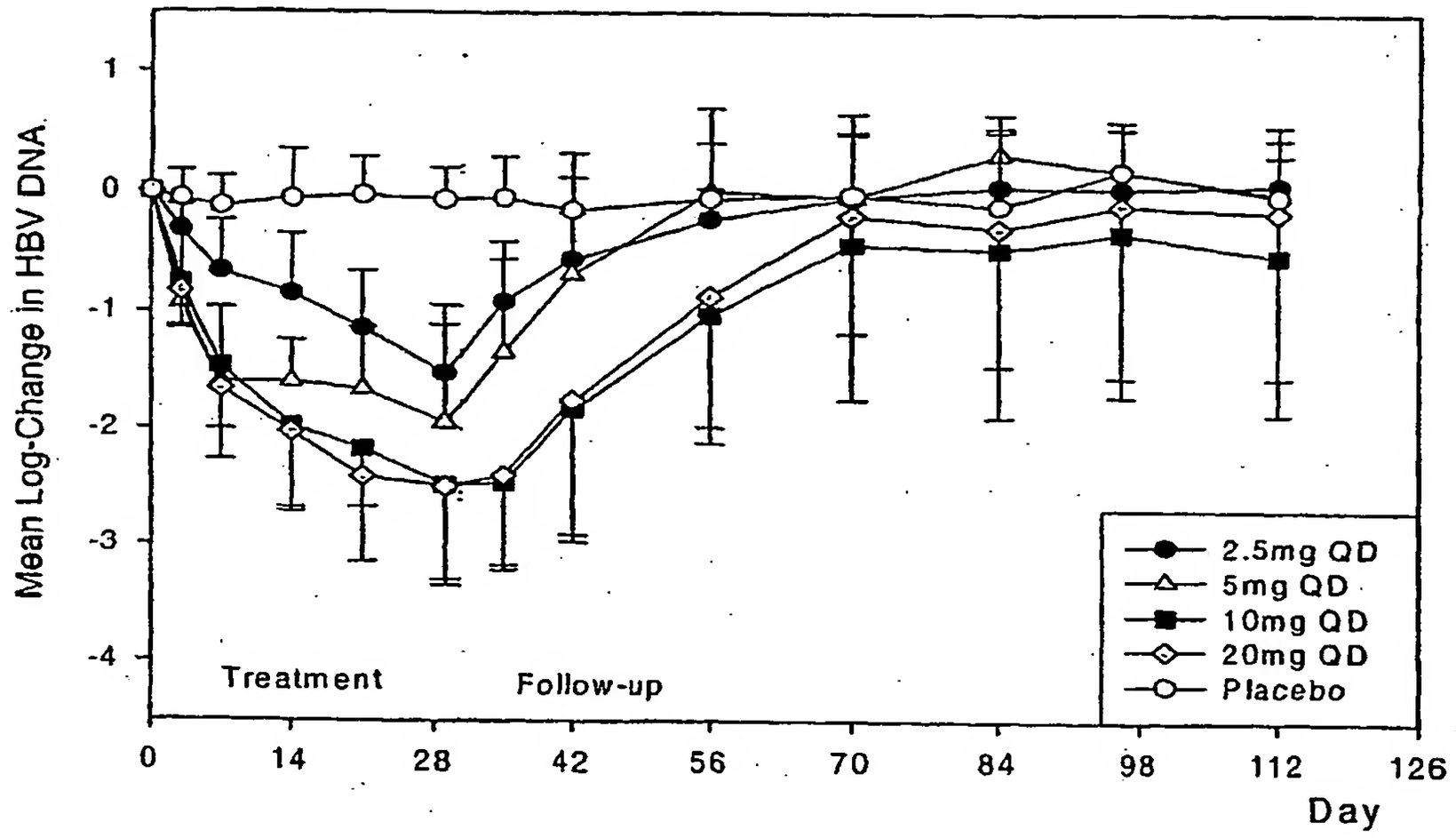

Mean Log-Change in HBV DNA
1
0
-1
-2
-3
-4
Treatment
Follow-up
0
14
28
42
56
70
84
98
112
126
Day
2.5mg QD
5mg QD
10mg QD
20mg QD
Placebo

FIGURE 2

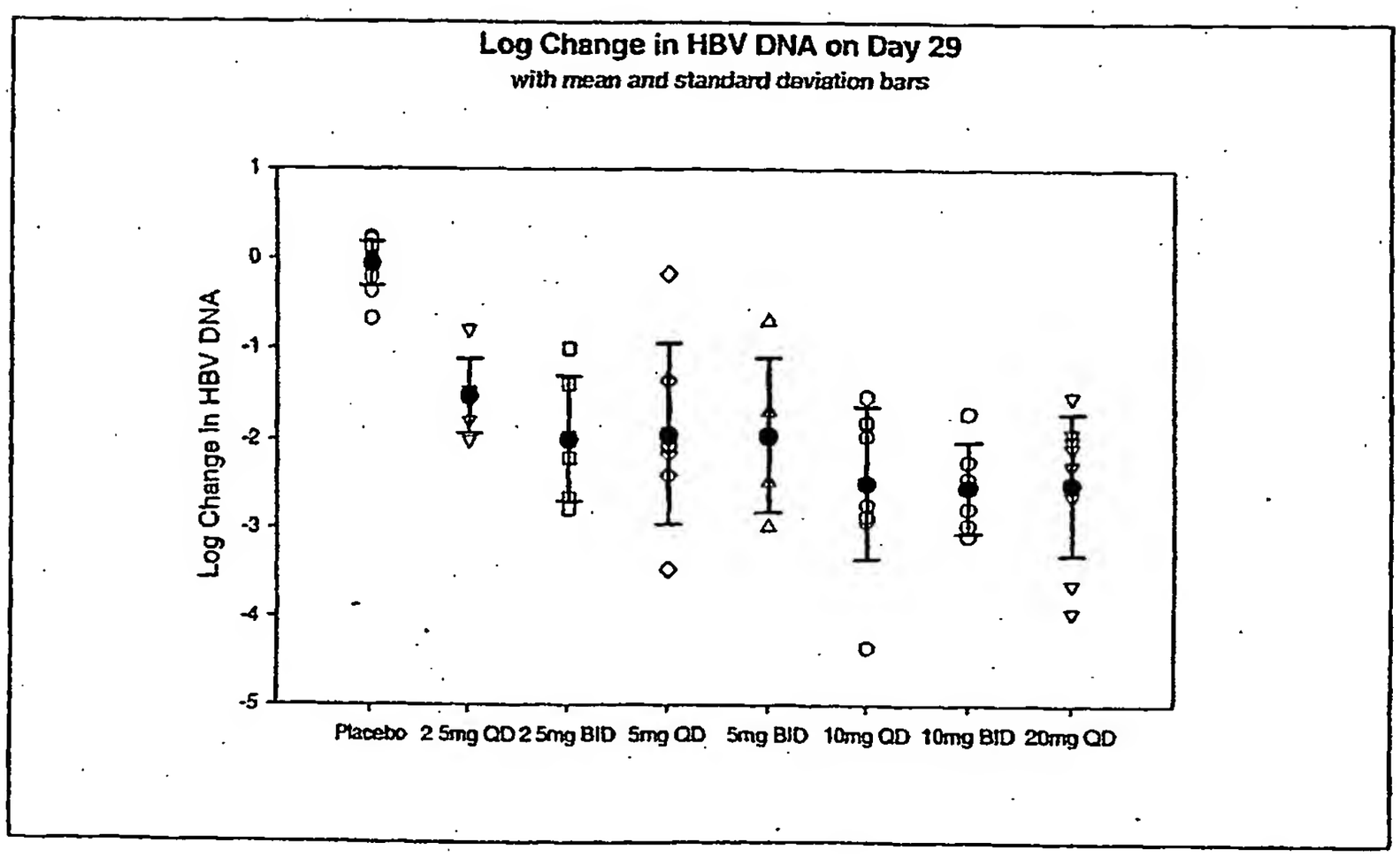
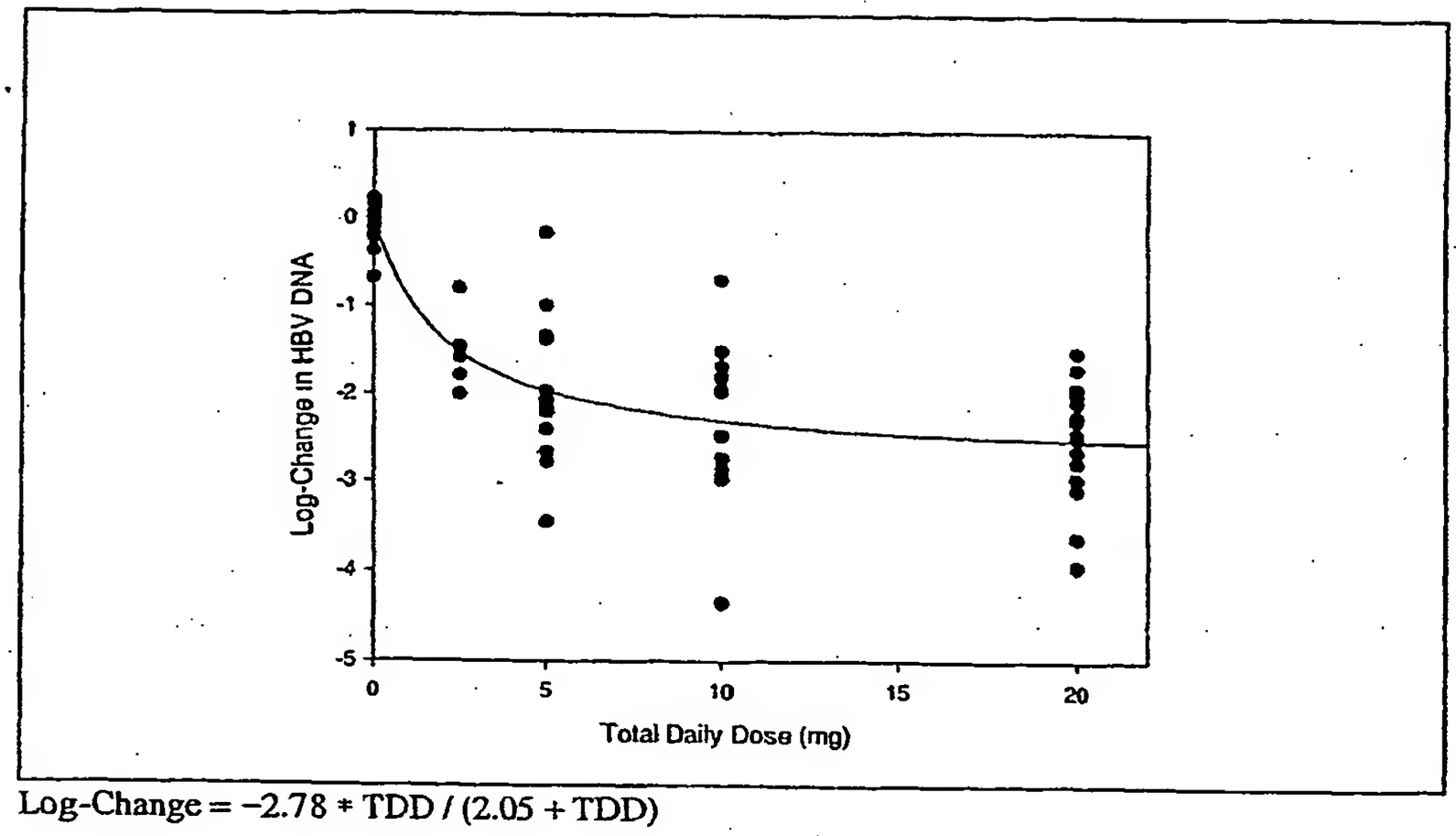
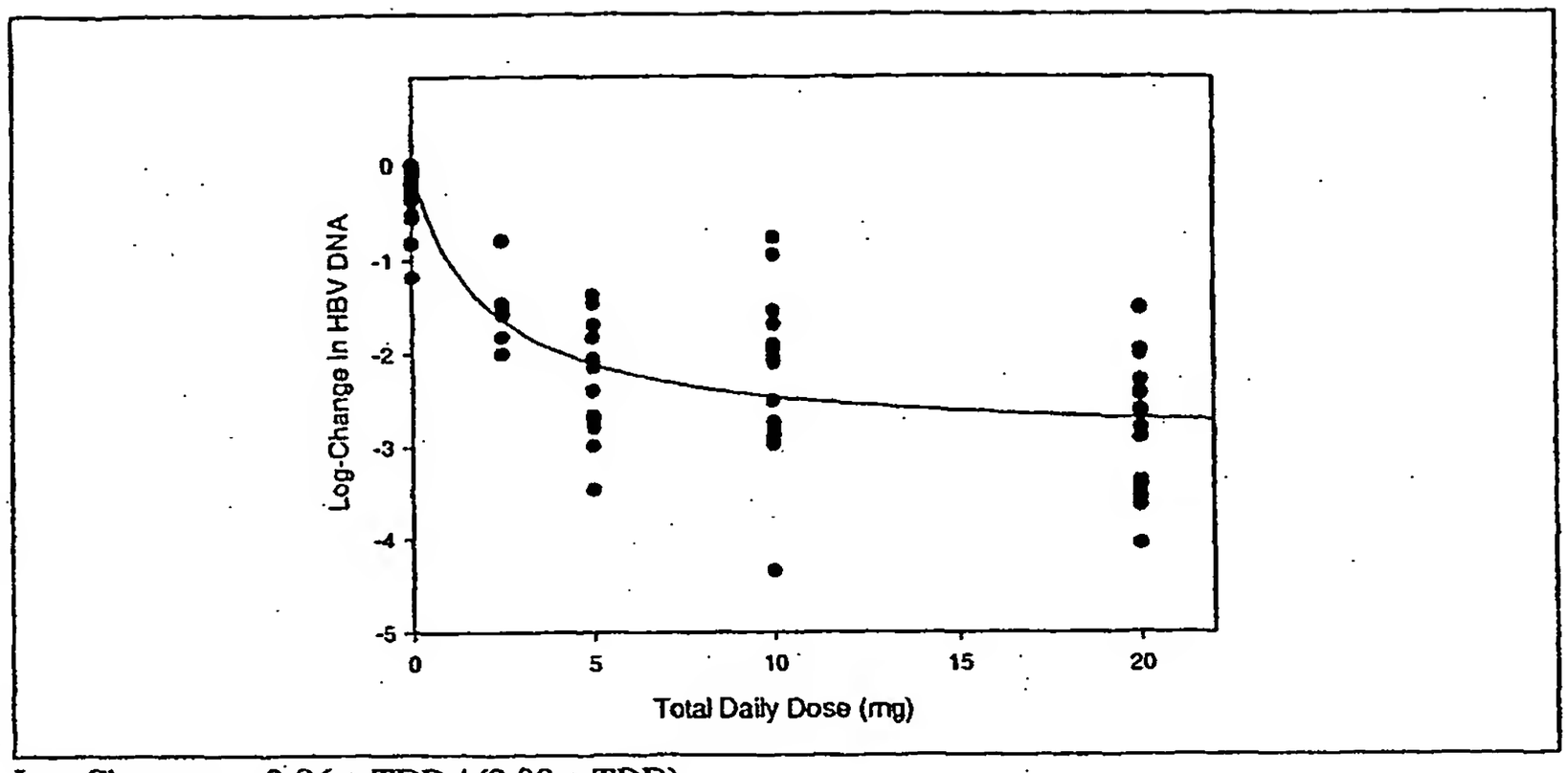

Log Change in HBV DNA on Day 29
with mean and standard deviation bars
Log Change In HBV DNA
1
0
-1
-2
-3
-4
-5
Placebo
2.5mg QD
2.5mg BID
5mg QD
5mg BID
10mg QD
10mg BID
20mg QD

FIGURE 3



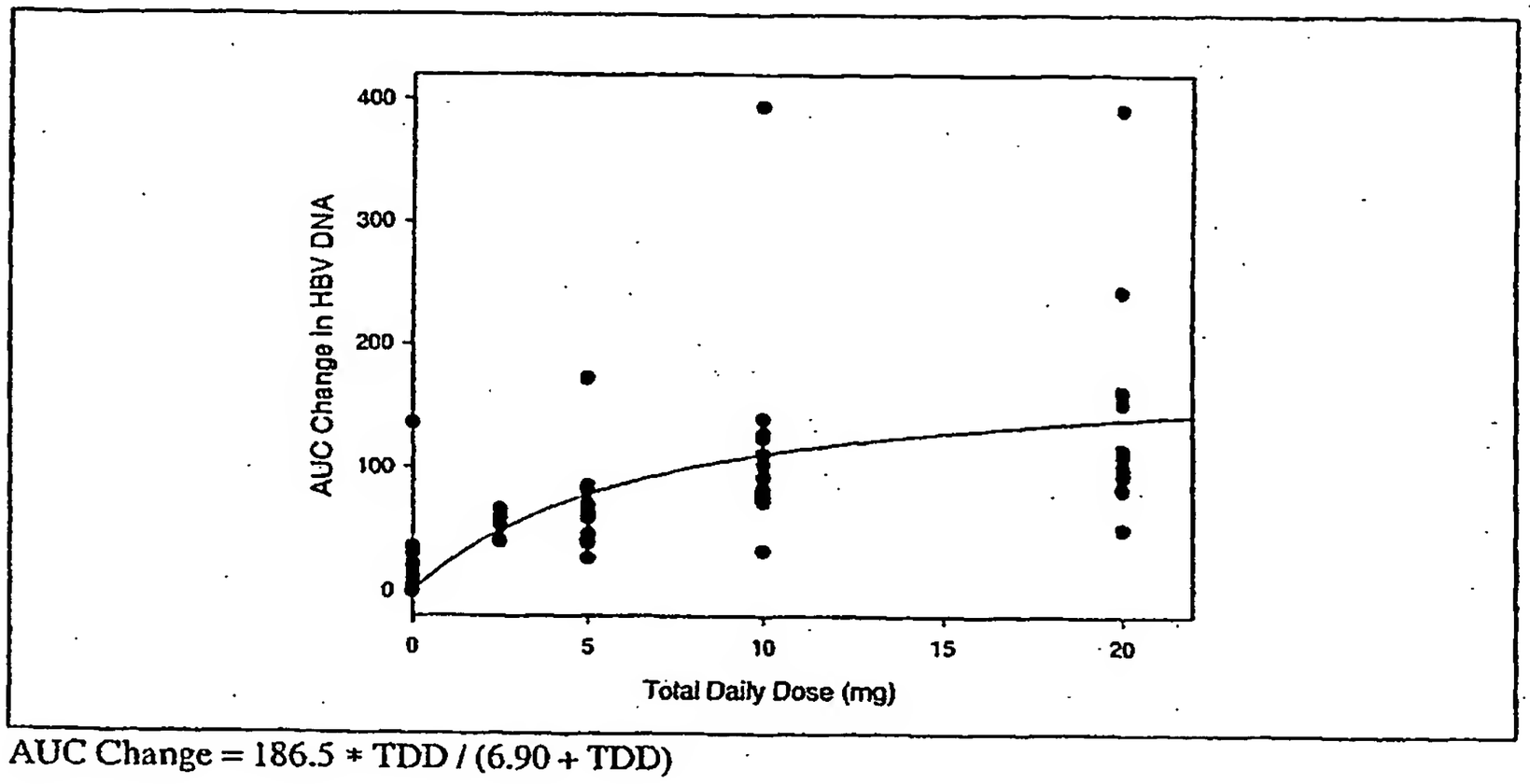
Log-Change = −2.78 * TDD / (2.05 + TDD)

FIGURE 4



Log-Change = −2.96 * TDD / (2.00 + TDD)

FIGURE 5



AUC Change = 186.5 * TDD / (6.90 + TDD)

FIGURE 6

INTERNATIONAL SEARCH REPORT

Int nal Application No
PCT/US 02/33641

A. CLASSIFICATION OF SUBJECT MATTER
IPC 7 C07F9/6561 A61K31/675

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
IPC 7 C07F A61K

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)
EPO-Internal, BIOSIS

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	US 5 840 716 A (TAKASHIMA HIDEAKI ET AL) 24 November 1998 (1998-11-24)	1-8, 10-16
Y	claims 1,16-18 column 60, line 40 - line 41 column 61, line 8 - line 11	1-16
Y	EP 0 919 562 A (MITSUBISHI CHEM CORP) 2 June 1999 (1999-06-02) claims 1,4,5 page 44, paragraph '0052!	1-16
Y	EP 0 632 048 A (MITSUBISHI CHEM IND) 4 January 1995 (1995-01-04) claims 1,3;12-14	1-16

-/--

[X] Further documents are listed in the continuation of box C.

[X] Patent family members are listed in annex.

° Special categories of cited documents :

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier document but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.

"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
21 February 2003	06/03/2003

Name and mailing address of the ISA	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Tx. 31 651 epo nl, Fax (+31-70) 340-3016	Beranová, P

Form PCT/ISA/210 (second sheet) (July 1992)

INTERNATIONAL SEARCH REPORT

Int nal Application No
PCT/US 02/33641

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication,where appropriate, of the relevant passages	Relevant to claim No.
Y	US 5 977 061 A (DE CLERCQ ERIK DESIRE ALICE ET AL) 2 November 1999 (1999-11-02) claim 1 column 13, line 27 - line 28	1-16
Y	EP 0 832 896 A (MITSUBISHI CHEM CORP) 1 April 1998 (1998-04-01) claims 1,12,13 page 30, line 17	1-16
Y	YOKOTA T ET AL: "INHIBITORY EFFECTS OF PHOSPHONATE NUCLEOTIDE ANALOGUES ON HUMAN HEPATITIS B VIRUS DNA SYNTHESIS" ANTIVIRAL RESEARCH, vol. 17, no. SUPPL. 1, 1992, page 146 XP008014106 ISSN: 0166-3542 Abstract	1-16

INTERNATIONAL SEARCH REPORT

ıational application No.
PCT/US 02/33641

Box I Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)

This International Search Report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. [X] Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:

Although claims 7 – 16 are directed to a method of treatment of the human/animal body, the search has been carried out and based on the alleged effects of the compound/composition.

2. [ ] Claims Nos.:
because they relate to parts of the International Application that do not comply with the prescribed requirements to such an extent that no meaningful International Search can be carried out, specifically:

3. [ ] Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

1. [ ] As all required additional search fees were timely paid by the applicant, this International Search Report covers all searchable claims.

2. [ ] As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.

3. [ ] As only some of the required additional search fees were timely paid by the applicant, this International Search Report covers only those claims for which fees were paid, specifically claims Nos.:

4. [ ] No required additional search fees were timely paid by the applicant. Consequently, this International Search Report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

[ ] The additional search fees were accompanied by the applicant's protest.

[ ] No protest accompanied the payment of additional search fees.

INTERNATIONAL SEARCH REPORT

Ir onal Application No
PCT/US 02/33641

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
US 5840716 A	24-11-1998	AT 199907 T	15-04-2001
		CA 2195262 A1	19-07-1997
		CN 1164534 A ,B	12-11-1997
		DE 69704286 D1	26-04-2001
		DE 69704286 T2	25-10-2001
		DK 785208 T3	23-04-2001
		EP 0785208 A1	23-07-1997
		ES 2155219 T3	01-05-2001
		GR 3035776 T3	31-07-2001
		JP 3148139 B2	19-03-2001
		JP 9255695 A	30-09-1997
		PT 785208 T	29-06-2001
EP 0919562 A	02-06-1999	DE 69716924 D1	12-12-2002
		EP 0919562 A1	02-06-1999
		US 6194398 B1	27-02-2001
		CN 1232466 A ,B	20-10-1999
		WO 9806726 A1	19-02-1998
		KR 2000029952 A	25-05-2000
EP 0632048 A	04-01-1995	AT 199906 T	15-04-2001
		CA 2126601 A1	30-12-1994
		CN 1106017 A ,B	02-08-1995
		DE 69426904 D1	26-04-2001
		DE 69426904 T2	11-10-2001
		EP 0632048 A1	04-01-1995
		JP 7070159 A	14-03-1995
		US 6037335 A	14-03-2000
US 5977061 A	02-11-1999	AU 5268696 A	07-11-1996
		WO 9633200 A1	24-10-1996
		EP 0821690 A1	04-02-1998
		JP 11511114 T	28-09-1999
EP 0832896 A	01-04-1998	EP 0832896 A1	01-04-1998
		US 6197775 B1	06-03-2001
		CA 2224710 A1	03-01-1997
		CN 1192744 A ,B	09-09-1998
		WO 9700262 A1	03-01-1997